



US-102

SEQUENCE LISTING

<110> Ajinomoto Co. Inc.

<120> Method for Producing L-Amino Acid Using Methylotroph

<130> OP1627/US-102

<140>

<141> 2003-11-

<150> JP 2002-336315

<151> 2002-11-20

<160> 14

<170> PatentIn Ver. 2.0

<210> 1

<211> 711

<212> DNA

<213> Brevibacterium lactofermentum

<220>

<221> CDS

<222> (1)..(711)

<400> 1

atg gtg atc atg gaa atc ttc att aca ggt ctg ctt ttg ggg gcc agt 48
Met Val Ile Met Glu Ile Phe Ile Thr Gly Leu Leu Leu Gly Ala Ser
1 5 10 15

ctt tta ctg tcc atc gga ccg cag aat gta ctg gtg att aaa caa gga 96
Leu Leu Leu Ser Ile Gly Pro Gln Asn Val Leu Val Ile Lys Gln Gly
20 25 30

att aag cgc gaa gga ctc att gcg gtt ctt ctc gtg tgt tta att tct 144
Ile Lys Arg Glu Gly Leu Ile Ala Val Leu Leu Val Cys Leu Ile Ser
35 40 45

gac gtc ttt ttg ttc atc gcc ggc acc ttg ggc gtt gat ctt ttg tcc 192
Asp Val Phe Leu Phe Ile Ala Gly Thr Leu Gly Val Asp Leu Leu Ser
50 55 60

aat gcc gcg ccg atc gtg ctc gat att atg cgc tgg ggt ggc atc gct 240
Asn Ala Ala Pro Ile Val Leu Asp Ile Met Arg Trp Gly Gly Ile Ala
65 70 75 80

tac ctg tta tgg ttt gcc gtc atg gca gcg aaa gac gcc atg aca aac 288
Tyr Leu Leu Trp Phe Ala Val Met Ala Ala Lys Asp Ala Met Thr Asn
85 90 95

aag gtg gaa gcg cca cag atc att gaa gaa aca gaa cca acc gtg ccc 336
Lys Val Glu Ala Pro Gln Ile Ile Glu Glu Thr Glu Pro Thr Val Pro
100 105 110

gat gac acg cct ttg ggc ggt tcg gcg gtg gcc act gac acg cgc aac 384
Asp Asp Thr Pro Leu Gly Gly Ser Ala Val Ala Thr Asp Thr Arg Asn
115 120 125

cgg gtg cgg gtg gag gtg agc gtc gat aag cag cgg gtt tgg gta aag 432
Arg Val Arg Val Glu Val Ser Val Asp Lys Gln Arg Val Trp Val Lys
130 135 140

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| | |
|---|-----|
| ccc atg ttg atg gca atc gtg ctg acc tgg ttg aac ccg aat gcg tat | 480 |
| Pro Met Leu Met Ala Ile Val Leu Thr Trp Leu Asn Pro Asn Ala Tyr | |
| 145 150 155 160 | |
| ttg gac gcg ttt gtg ttt atc ggc ggc gtc ggc gcg caa tac ggc gac | 528 |
| Leu Asp Ala Phe Val Phe Ile Gly Gly Val Gly Ala Gln Tyr Gly Asp | |
| 165 170 175 | |
| acc gga cgg tgg att ttc gcc gct ggc gcg ttc gcg gca agc ctg atc | 576 |
| Thr Gly Arg Trp Ile Phe Ala Ala Gly Ala Phe Ala Ala Ser Leu Ile | |
| 180 185 190 | |
| tgg ttc ccg ctg gtg ggt ttc ggc gca gca ttg tca cgc ccg ctg | 624 |
| Trp Phe Pro Leu Val Gly Phe Gly Ala Ala Ala Leu Ser Arg Pro Leu | |
| 195 200 205 | |
| tcc agc ccc aag gtg tgg cgc tgg atc aac gtc gtc gtg gca gtt gtg | 672 |
| Ser Ser Pro Lys Val Trp Arg Trp Ile Asn Val Val Val Ala Val Val | |
| 210 215 220 | |
| atg acc gca ttg gcc atc aaa ctg atg ttg atg ggt tag | 711 |
| Met Thr Ala Leu Ala Ile Lys Leu Met Leu Met Gly | |
| 225 230 235 | |

<210> 2

<211> 236

<212> PRT

<213> *Brevibacterium lactofermentum*

<400> 2

| | |
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| Met Val Ile Met Glu Ile Phe Ile Thr Gly Leu Leu Leu Gly Ala Ser | |
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| Leu Leu Leu Ser Ile Gly Pro Gln Asn Val Leu Val Ile Lys Gln Gly | |
| 20 25 30 | |
| Ile Lys Arg Glu Gly Leu Ile Ala Val Leu Leu Val Cys Leu Ile Ser | |
| 35 40 45 | |
| Asp Val Phe Leu Phe Ile Ala Gly Thr Leu Gly Val Asp Leu Leu Ser | |
| 50 55 60 | |
| Asn Ala Ala Pro Ile Val Leu Asp Ile Met Arg Trp Gly Gly Ile Ala | |
| 65 70 75 80 | |
| Tyr Leu Leu Trp Phe Ala Val Met Ala Ala Lys Asp Ala Met Thr Asn | |
| 85 90 95 | |
| Lys Val Glu Ala Pro Gln Ile Ile Glu Glu Thr Glu Pro Thr Val Pro | |
| 100 105 110 | |
| Asp Asp Thr Pro Leu Gly Gly Ser Ala Val Ala Thr Asp Thr Arg Asn | |
| 115 120 125 | |
| Arg Val Arg Val Glu Val Ser Val Asp Lys Gln Arg Val Trp Val Lys | |
| 130 135 140 | |
| Pro Met Leu Met Ala Ile Val Leu Thr Trp Leu Asn Pro Asn Ala Tyr | |
| 145 150 155 160 | |
| Leu Asp Ala Phe Val Phe Ile Gly Gly Val Gly Ala Gln Tyr Gly Asp | |
| 165 170 175 | |
| Thr Gly Arg Trp Ile Phe Ala Ala Gly Ala Phe Ala Ala Ser Leu Ile | |
| 180 185 190 | |
| Trp Phe Pro Leu Val Gly Phe Gly Ala Ala Ala Leu Ser Arg Pro Leu | |
| 195 200 205 | |
| Ser Ser Pro Lys Val Trp Arg Trp Ile Asn Val Val Val Ala Val Val | |
| 210 215 220 | |
| Met Thr Ala Leu Ala Ile Lys Leu Met Leu Met Gly | |
| 225 230 235 | |

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<210> 3
 <211> 1197
 <212> DNA
 <213> Escherichia coli

<220>
 <221> CDS
 <222> (272)..(1153)

<400> 3
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 caattccacg gcgatcgca cccaaacgcag tgatcaccag ataatgtgtt gcgatgacag 120
 tgtcaactg gttattcctt taaggggtga gttgttctta agggaaagcat aaaaaaaaaaca 180
 tgcataacaac aatcagaacg gttctgtctg cttgcttta atgcccatacc aaacgtacca 240
 ttgagacact tgtttgacca gaggatggcc c atg ttc acg gga agt att gtc 292
 Met Phe Thr Gly Ser Ile Val
 1 5
 gcg att gtt act ccg atg gat gaa aaa ggt aat gtc ttt cgg gct agc 340
 Ala Ile Val Thr Pro Met Asp Glu Lys Gly Asn Val Cys Arg Ala Ser
 10 15 20
 ttg aaa aaa ctg att gat tat cat gtc gcc agc ggt act tcc gcg atc 388
 Leu Lys Lys Leu Ile Asp Tyr His Val Ala Ser Gly Thr Ser Ala Ile
 25 30 35
 gtt tct gtt ggc acc act ggc gag tcc gct acc tta aat cat gac gaa 436
 Val Ser Val Gly Thr Thr Gly Glu Ser Ala Thr Leu Asn His Asp Glu
 40 45 50 55
 cat gct gat gtg gtg atg atg acg ctg gat ctg gct gat ggg cgc att 484
 His Ala Asp Val Val Met Met Thr Leu Asp Leu Ala Asp Gly Arg Ile
 60 65 70
 ccg gta att gcc ggg acc ggc gct aac gct act gcg gaa gcc att agc 532
 Pro Val Ile Ala Gly Thr Gly Ala Asn Ala Thr Ala Glu Ala Ile Ser
 75 80 85
 ctg acg cag cgc ttc aat gac agt ggt atc gtc ggc tgc ctg acg gta 580
 Leu Thr Gln Arg Phe Asn Asp Ser Gly Ile Val Gly Cys Leu Thr Val
 90 95 100
 acc cct tac tac aat cgt ccg tcg caa gaa ggt ttg tat cag cat ttc 628
 Thr Pro Tyr Tyr Asn Arg Pro Ser Gln Glu Gly Leu Tyr Gln His Phe
 105 110 115
 aaa gcc atc gct gag cat act gac ctg ccg caa att ctg tat aat gtg 676
 Lys Ala Ile Ala Glu His Thr Asp Leu Pro Gln Ile Leu Tyr Asn Val
 120 125 130 135
 ccg tcc cgt act ggc tgc gat ctg ctc ccg gaa acg gtg ggc cgt ctg 724
 Pro Ser Arg Thr Gly Cys Asp Leu Leu Pro Glu Thr Val Gly Arg Leu
 140 145 150
 gcg aaa gta aaa aat att atc gga atc aaa gag gca aca ggg aac tta 772
 Ala Lys Val Lys Asn Ile Ile Gly Ile Lys Glu Ala Thr Gly Asn Leu
 155 160 165
 acg cgt gta aac cag atc aaa gag ctg gtt tca gat gat ttt gtt ctg 820
 Thr Arg Val Asn Gln Ile Lys Glu Leu Val Ser Asp Asp Phe Val Leu
 170 175 180
 ctg agc ggc gat gat gcg agc gcg ctg gac ttc atg caa ttg ggc ggt 868
 Leu Ser Gly Asp Asp Ala Ser Ala Leu Asp Phe Met Gln Leu Gly Gly
 185 190 195
 cat ggg gtt att tcc gtt acg act aac gtc gca gcg cgt gat atg gcc 916

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| | | | | | | | | | | | | | | | | |
|------------|------------|------------|------------|----------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|------|------|
| His | Gly | Val | Ile | Ser | Val | Thr | Thr | Asn | Val | Ala | Ala | Arg | Asp | Met | Ala | |
| 200 | | | | | 205 | | | | | 210 | | | | | 215 | |
| cag | atg | tgc | aaa | ctg | gca | gca | gaa | gaa | cat | ttt | gcc | gag | gca | cgc | gtt | 964 |
| Gln | Met | Cys | Lys | Leu | Ala | Ala | Glu | Glu | His | Phe | Ala | Glu | Ala | Arg | Val | |
| | | | | 220 | | | | | 225 | | | | | 230 | | |
| att | aat | cag | cgt | ctg | atg | cca | tta | cac | aac | aaa | cta | ttt | gtc | gaa | ccc | 1012 |
| Ile | Asn | Gln | Arg | Leu | Met | Pro | Leu | His | Asn | Lys | Leu | Phe | Val | Glu | Pro | |
| | | | | 235 | | | | | 240 | | | | | 245 | | |
| aat | cca | atc | ccg | gtg | aaa | tgg | gca | tgt | aag | gaa | ctg | ggt | ctt | gtg | gcg | 1060 |
| Asn | Pro | Ile | Pro | Val | Lys | Trp | Ala | Cys | Lys | Glu | Leu | Gly | Leu | Val | Ala | |
| | | | | 250 | | | | | 255 | | | | | 260 | | |
| acc | gat | acg | ctg | cgc | ctg | cca | atg | aca | cca | atc | acc | gac | agt | ggt | cgt | 1108 |
| Thr | Asp | Thr | Leu | Arg | Leu | Pro | Met | Thr | Pro | Ile | Thr | Asp | Ser | Gly | Arg | |
| | | | | 265 | | | | | 270 | | | | | 275 | | |
| gag | acg | gtc | aga | gca | gca | ctt | aag | cat | gcc | ggt | ttg | ctg | taa | | 1150 | |
| Glu | Thr | Val | Arg | Ala | Ala | Leu | Lys | His | Ala | Gly | Leu | Leu | | | | |
| | | | | 280 | | | | | 285 | | | | | 290 | | |
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<210> 4

<211> 292

<212> PRT

<213> Escherichia coli

<400> 4

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| Met | Phe | Thr | Gly | Ser | Ile | Val | Ala | Ile | Val | Thr | Pro | Met | Asp | Glu | Lys | |
| 1 | | | | | 5 | | | | 10 | | | | | 15 | | |
| Gly | Asn | Val | Cys | Arg | Ala | Ser | Leu | Lys | Lys | Leu | Ile | Asp | Tyr | His | Val | |
| | | | | | 20 | | | | 25 | | | | | 30 | | |
| Ala | Ser | Gly | Thr | Ser | Ala | Ile | Val | Ser | Val | Gly | Thr | Thr | Gly | Glu | Ser | |
| | | | | | 35 | | | | 40 | | | | | 45 | | |
| Ala | Thr | Leu | Asn | His | Asp | Glu | His | Ala | Asp | Val | Val | Met | Met | Thr | Leu | |
| | | | | | 50 | | | | 55 | | | | | 60 | | |
| Asp | Leu | Ala | Asp | Gly | Arg | Ile | Pro | Val | Ile | Ala | Gly | Thr | Gly | Ala | Asn | |
| | | | | | 65 | | | | 70 | | | | | 75 | | 80 |
| Ala | Thr | Ala | Glu | Ala | Ile | Ser | Leu | Thr | Gln | Arg | Phe | Asn | Asp | Ser | Gly | |
| | | | | | 85 | | | | 90 | | | | | 95 | | |
| Ile | Val | Gly | Cys | Leu | Thr | Val | Thr | Pro | Tyr | Tyr | Asn | Arg | Pro | Ser | Gln | |
| | | | | | 100 | | | | 105 | | | | | 110 | | |
| Glu | Gly | Leu | Tyr | Gln | His | Phe | Lys | Ala | Ile | Ala | Glu | His | Thr | Asp | Leu | |
| | | | | | 115 | | | | 120 | | | | | 125 | | |
| Pro | Gln | Ile | Leu | Tyr | Asn | Val | Pro | Ser | Arg | Thr | Gly | Cys | Asp | Leu | Leu | |
| | | | | | 130 | | | | 135 | | | | | 140 | | |
| Pro | Glu | Thr | Val | Gly | Arg | Leu | Ala | Lys | Val | Lys | Asn | Ile | Ile | Gly | Ile | |
| | | | | | 145 | | | | 150 | | | | | 155 | | 160 |
| Lys | Glu | Ala | Thr | Gly | Asn | Leu | Thr | Arg | Val | Asn | Gln | Ile | Lys | Glu | Leu | |
| | | | | | 165 | | | | 170 | | | | | 175 | | |
| Val | Ser | Asp | Asp | Phe | Val | Leu | Leu | Ser | Gly | Asp | Asp | Ala | Ser | Ala | Leu | |
| | | | | | 180 | | | | 185 | | | | | 190 | | |
| Asp | Phe | Met | Gln | Leu | Gly | Gly | His | Gly | Val | Ile | Ser | Val | Thr | Thr | Asn | |
| | | | | | 195 | | | | 200 | | | | | 205 | | |
| Val | Ala | Ala | Arg | Asp | Met | Ala | Gln | Met | Cys | Lys | Leu | Ala | Ala | Glu | Glu | |
| | | | | | 210 | | | | 215 | | | | | 220 | | |
| His | Phe | Ala | Glu | Ala | Arg | Val | Ile | Asn | Gln | Arg | Leu | Met | Pro | Leu | His | |
| | | | | | 225 | | | | 230 | | | | | 235 | | 240 |

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Asn Lys Leu Phe Val Glu Pro Asn Pro Ile Pro Val Lys Trp Ala Cys
245 250 255
Lys Glu Leu Gly Leu Val Ala Thr Asp Thr Leu Arg Leu Pro Met Thr
260 265 270
Pro Ile Thr Asp Ser Gly Arg Glu Thr Val Arg Ala Ala Leu Lys His
275 280 285
Ala Gly Leu Leu
290

<210> 5
<211> 36
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 5
cggttggagg tgagcgtcgg taagcagcgg gtttgg 36

<210> 6
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 6
gtcttttgt tcatcaccag caccttggc gtt 33

<210> 7
<211> 39
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 7
aggaaattcc ccgttctgga taatgtttt tgcgccgac 39

<210> 8
<211> 58
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 8
cgatgcattc tagatctaactgcagggtg aaattgttat ccgctcacaa ttccacac 58

<210> 9
<211> 64

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<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 9
catttcctgc aggcaaagga gatgagcgta atgggtatca tggaaatctt cattacaggt 60
ctgc 64

<210> 10
<211> 50
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 10
gggcgagcta gaagagctcc aaaacccgcg aaaactaacc catcaacatc 50

<210> 11
<211> 35
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 11
tgacctgcag gtttgcacag aggatggccc atgtt 35

<210> 12
<211> 36
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 12
cattcttagat ccctaaactt tacagcaaac cggcat 36

<210> 13
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 13
gcacggatca ctgtattcggt ctgcaacttt 30

<210> 14

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<211> 32
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: primer

<400> 14
gccgttgtc taggatggtt gttcttggat ca

32